STUDIES ON THE SUSCEPTIBILITY

Level Of The Housefly Musca Domestica Nebulo To DDT

IN ALIGARH

PROJECT REPORT SUBMITTED IN THE PARTIAL FULFILMENT
OF THE REQUIREMENTS FOR THE AWARD OF THE
DEGREE OF MASTER OF SCIENCE IN ZOOLOGY (PARASITOLOGY).

Course No. 5-Zy-72

1975—76

BY

JOSEPHINE PONDAYI—ZESAGULI (MRS)

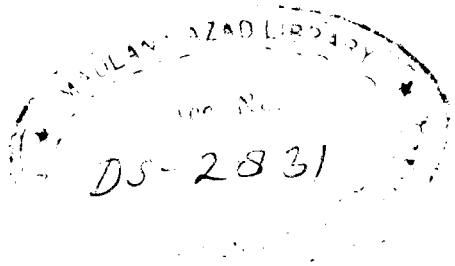
M.Sc. (Final)

ROLL No. 71 ZYM—32

ENROLMENT No. L 4111

DEPARTMENT OF ZOOLOGY
ALIGARH MUSLIM UNIVERSITY
ALIGARH

JUNE, 1976



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ACKNOWLEDGEMENTS

I am grateful to Dr. Jamil A. Ansari for supervising my project. I also wish to thank Professor S. M. Alam, Head of the Zoology Department, Aligarh Muslim University, Aligarh for providing the necessary laboratory facilities. Lastly, but not the least, I wish to express my gratitude to the Government of India, Cultural Scholarship which financed my studies.

JOSEPHINE PONDAYI ZESAGULI (MRS)
Section of Parasitology,
Zoology Department,
Aligarh Muslim University,
ALIGARH.

CONTENTS

INTRODUCTION

The housefly, Musca nebula is a cosmopolitan species and has great medical importance. The fly serves as a direct agent and vector of many pathogens especially those causing enteric diseases. These enteric diseases, for example, dysentry are due to the fact that flies frequent toilets, dumps and even faeces for egg-laying. Here, they pick parasitic protozoans e.g. Ent-amoeba, Giardia etc., and helminth ova on their hairy and sticky feet or actually swallow them. Later, they deposit the organisms in kitchens, dining tables and exposed edible substances. Due to their regurgitation habit, the flies vomit the germs from their food reservoir with the saliva to dissolve the sugar and also via their faeces (fly specks). Chandler reports that such specks are said to be produced by a well-fed fly every 5 minutes all day. So, flies in this way transmit bacteria causing anthrax, typhoid, dysentry and food poisoning. They also harbour the virus for poliomyelitis and may produce myiasis. Spirochaetes causing yaws are also transmitted by flies.

Obviously, the potential danger of flies cannot be minimised. The danger described above lies mainly in their feeding habits. It is also attributable to their high fecundity, fertility and the short duration of their developmental cycle. The fecundity is better appreciated if one realises that a female, within its life time which averages from 6 to 8 weeks, will have deposited up to 2,000 eggs. Hence, the danger of flies is multiplied in geometric progression.

Back home in Rhodesia, there are still many deaths and diseases among man and his livestock caused by parasites. Typhoid and dysentry are common among children whose lives are nipped in the bud and thus their potential is a great loss to the country's progress.

Man has since become aware of the imminent danger of flies and has tried to exterminate them mechanically and biologically. But the most effective measure is the control by chemicals so called as insecticides. The chemicals may include chlorinated hydrocarbons, organophosphorous compounds and plant products as pyrethroids. Among the chlorinated hydrocarbons, the most commonly used insecticides against flies is DDT.

The present project on susceptibility of houseflies to DDT in Aligarh was taken up so that I get exposed to the method of approach in the control of insects. This will make me better equipped to go back and help alleviate the problem of these pests which are of medical and veterinary importance together with other researches related to toxicology.

REVIEW OF LITERATURE

DDT was the first insecticide of the chlorinated hydrocarbon group to be used on a world wide scale against houseflies and other pests. This was also the first chemical to induce resistance in insects. Naturally it drew greater research effort than any other compound.

DDT was first made in 1874. Its insecticidal properties were discovered by Muller in the Basle Laboratories of the Swiss Company of J.R. Geigy S.A. in 1939. The new insecticide quickly proved exceptionally effective by checking the plague of Colorado beetle in 1941 which threatened the Swiss potato crop at a time when all food crops were of utmost importance. The insecticide was thoroughly tested against a wide range of insects including mosquitoes and houseflies. It took a year to establish its insecticidal action.

The difficulties of communication during World War II restricted the spread of this information to the free world. The chemical was ultimately introduced into the United Kingdom and the U.S.A. towards the end of 1942, and by that time much was known concerning the compound.

Muller (1939) reported that compounding DDT into 5 or 10% dusts and sprays proved to have a lasting effect against flies and mosquitoes in rooms, barracks and farm buildings.

Maisman (1944) showed that DDT had effective residual action and acted as a nerve poison on the flies.

Buxton (1945) described the work carried out by Busvine (1945) showing that the lethal dose of DDT for adult Musca domestica was 1 mg per sq. cm. of surface. Work has also been done on the toxic manifestation of DDT in other animals including man.

Lindquist and co-workers (1944) reported that DDT applied as a spray in a suitable solvent remained as a nearly invisible deposit after the liquid had volatilised and acted as a residual contact insecticide against houseflies. Comparisons of the insecticidal results of DDT in various compounds as paints,

waxes etc. have also been done.

Sweetman (1946) recommended spot treatment of DDT where flies collect for feeding and resting. In 1947 Sweetman got excellent fly control with DDT in two dairy herds and respective barns.

Keller (1955) tested the effectiveness of DDT in baits.

As more and more DDT was used it was noticed that there was gradual decrease in its effectiveness. This was because resistance had developed in the flies which must not be confused with vigor tolerance (Hoskins and Gordon, 1956). There are two types of resistance which have been differentiated as behavioral and physiological (genetic) resistances.

Hess (1952) was the first to describe behavioral resistance in mosquitoes following contact with DDT. Since then it has been noticed in flies also.

Busvine (1963) observed that vigor tolerance and behavioral resistance were not likely to present serious obstacles as physiological resistance to major insect control schemes.

Physiological resistance is due to the presence of an enzyme DDT-hydrochlorinase which converts DDT to DDE. Perry and Hoskins (1951) found this detoxification of DDT as a factor in the resitance of houseflies. Linquist et. al. (1951) reported that this conversion of DDT to DDE is mainly concentrated in the fat body and gut region or in the cuticle hypodermis.

Whereas DDT is toxic, DDE is non-toxic to the insect. Hence, physiological resistance can develop to the point of immunity.

Wiesman (1957) observed that DDT-resistant flies had a lipoid barrier which dissolves the DDT. Hence, DDT becomes ineffective to such DDT-resistant flies. Brown (1961) recorded 36 instances of DDT-resistance.

Synthesis of enzymes is genetically controlled. Hence, DDT-resistant flies will also produce DDT-resistant progeny. The inheritence mechanism of DDT-resistance has been found to be monofactorial. Ansari (1969) agrees with this single gene hypothesis but found it to be not completely dominant.

Khan and Ansari (1964) noted that while Musca domestica domestica had developed resistance for DDT, the Indian housefly (Musca domestica nebulo) had not attained any significant degree of resistance. They also found that Musca domestica vicina got from Calcutta was liable to become more resistant than the nebulo sub-species..

Pal (1951) obtained only 1.6 - 2.0 times DDT-resistance in Musca domestica nebulo from a Delhi village.

Since it was first discovered in Sweden in (1946), resistance has been growing at an alarming rate and yet insecticides are still being effective. Busvine (1963) explains this observation as being due to the localisation of resistance in many instances and also because a limited number of species show resistance to both groups of chlorinated insecticides.

However if selection pressure continues, resistance will ultimately spread through the range of a species. This, I think, is a very important and crucial point to bear in mind in any toxicological undertaking since the rate of growth of resitance is well correlated with selective pressure.

MATERIALS AND TECHNIQUE

TEST FLIES:-

Flies were collected from various localities in Aligarh viz: Shamshad Market, S.N. Hall Mess and the City. These flies were pooled together to get a homogeneous stock.

The flies were maintained in wire net cages in the laboratory at room temperature. These were fed on buffalo milk which was diluted with water. The milk was put in petri dishes with soaked cotton. The petri dishes were changed daily.

REARING:-

The flies readily oviposited in the petri dishes. A batch of egges was seperately embedded in glass jars 4 X 8" under the pads of milk-soaked-cotton. Care was taken to embed a single batch of eggs in a jar, otherwise overcrowding would result in very small pupae and flies with a high mortality. The tops of the jars were covered with muslin cloths secured with a rubber band to prevent oviposition from outside flies and at the same time allow constant aeration. Everyday fresh soaked cotton was added to the jars and a dry layer of cotton was placed on the top of the pad on the fifth day. Larvae migrate to this dry layer and pupate in it.

The pupae were isolated from the dry cotton and put in petri dishes separetely according to the embedding date. These were allowed to hatch in muslin cloth cages. It took approximately 12 - 14 days from the initial stage of the egg to the emergence of adults. When these flies become four-day old, they are fully mature and are ready for testing.

T E S T I N G.

Pure DDT was taken as the test chemical. A stock solution of 1% concentration was made. At least five formulations ranging from 0.3125% to 1% were prepared. Dose of the insecticide was maintained as 0.0018 ml/fly.

Testing was done by topical application of the insecticide through a micro-applicator outfit. The flies were anaesthetized with carbon dioxide. A drop of the insecticide was applied on the dorsum of the flies.

Treated fliesvere dropped into tissue paper cages. Sugar cubes were provided as source of food and the opening of the cage was capped with dampened cotton. The cages were kept in the laboratory at room temperature.

After 24 hours of testing, the number of dead and live flies were counted in each cage. Mortality percentages were calculated for each DDT concentration using the formula:-

$$\frac{\text{Number of dead flies}}{\text{Total number of flies tested}} \times \frac{100}{1}$$

Live flies were those which could still fly and walk properly, the rest were considered as dead.

RESULTS AND DISCUSSION

From the Table the males seem to be more susceptible to DDT treatment than the females since there were more dead males (684) than females (384). This gives a ration of females dead to makes dead of 1 : 1.78.

It appears that females may be having a better inherent capacity for DDT detoxification than the males. But actually the higher percentage in males is because of their increased metabolic activity. Obviously rapid diffusion and absorption of this insecticide is taking place in males as compared to females. The results are very consistent and it may be concluded that males are more susceptible than females.

LC_{50} i.e. that concentration of DDT which is capable of killing 50% of the fly population as calculated from the graph was found to be 0.135. This is a lower value than what Khan and Ansari (1964) got for Aligarh flies. They found LC_{50} for Musca domestica nebulo to be 0.32 and that of Musca domestica vicina was 0.29 against DDT dissolved in Acetone.

Sen (1959) obtained 50% mortality with 4% DDT in Musca domestica vicina from Calcutta. While developing resistance to DDT in these flies, Khan and Ansari raised the value of the LC_{50} to 1.44 in the nebulo F_{16} and to 14.5 in the vicina F_{12} generations.

From this comparison even though it is admittedly not extensive, it can be concluded that Aligarh flies are still very susceptible to DDT treatment. This means that DDT could be used to control these flies. In fact Aligarh flies are such a menace that chemo-treatment should be tried with DDT but only for a short duration before the flies become resistant. A campaign must be launched to educate the people all around about the dangers of houseflies and how each individual can assist by proper sanitation and descriminate disposal of garbage and other refuse. This will get rid of the potential breeding places.

T A B L E

PERCENTAGE MORTALITY OF HOUSEFLIES AT DIFFERENT CONCENTRATIONS OF DDT

Concentration	No of flies tested	No.of flies dead			% Mortality
		Males	Females	Total	
0.03125	316	40	27	67	21.2
0.0625	342	68	51	119	34.7
0.125	332	117	57	174	52.4
0.25	313	121	72	193	61.68
0.5	316	164	76	240	75.3
1.0	320	174	98	272	85.0

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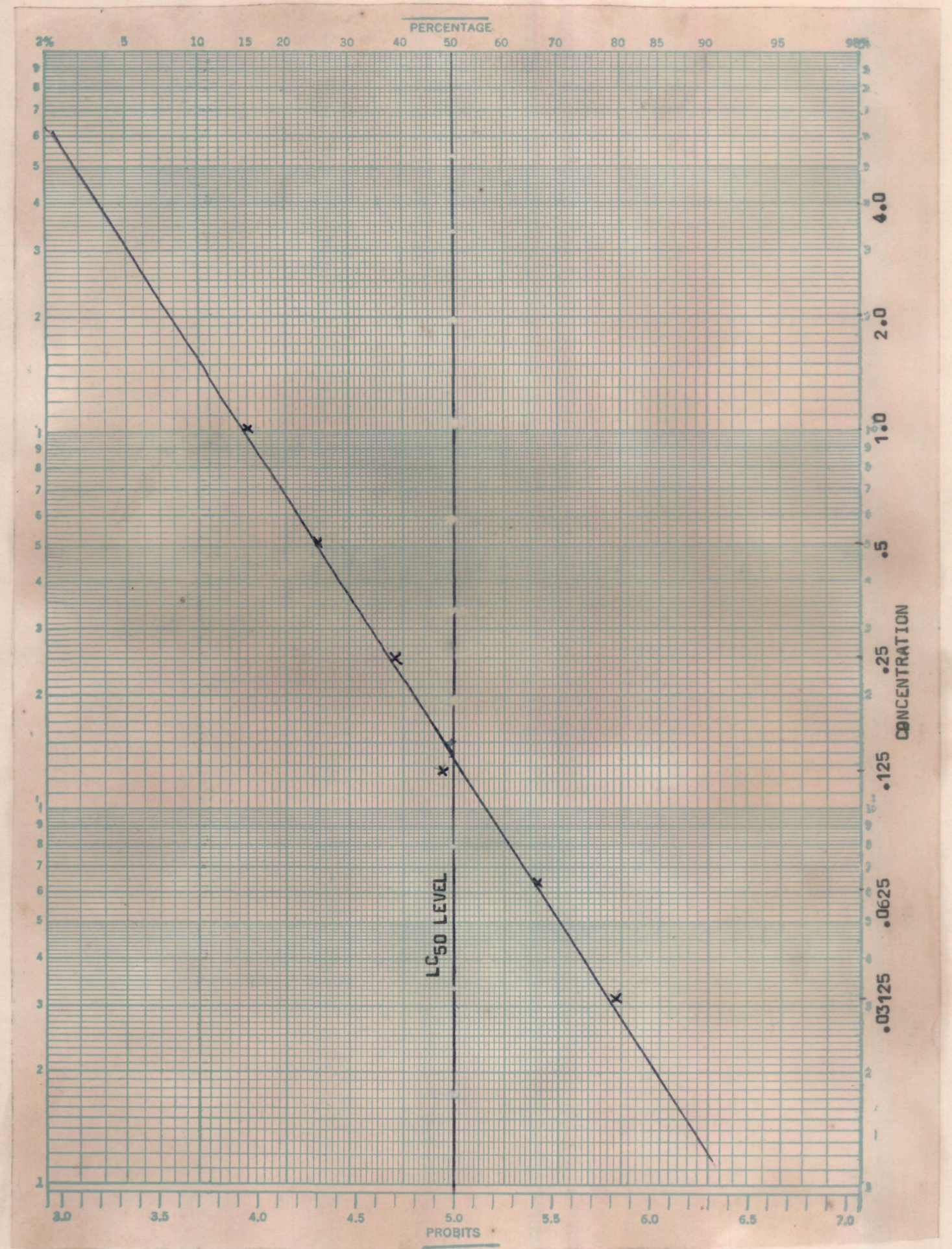
PERCENTAGE
2%
5
10
15
20
30
40
50
60
70
80
85
90
95
98%
4.0
2.0
1.0
.5
.25
.125
.0625
.03125
CONCENTRATION
LC50 LEVEL
3.0
3.5
4.0
4.5
5.0
5.5
6.0
6.5
7.0
PROBITS

100% mortality in the flies could be obtained by application of 7% DDT as calculated from the graph. This would, however, be impractical since acetone can dissolve only up to 6% DDT. Beyond this dosage the DDT will remain undissolved. So, another better solvent would have to be substituted for the acetone at this level of DDT concentration.

Therefore, females showed less susceptibility to DDT than males. LC_{50} was found to be only 0.135 which indicates that the flies have not yet developed resistance against DDT and 7% DDT is what is needed to give 100% mortality dissolved in some suitable solvent.

SUMMARY

Aligarh houseflies were collected from various localities and pooled to get a homogeneous stock. From this stock F_1 generation was obtained. Four-day old flies were tested by topical application on the dorsum with various concentrations of DDT dissolved in acetone. The susceptibility level of these flies was assessed from a plotted probability curve.

It was found that LC_{50} was 0.135 of DDT and that 7% DDT can give 100% mortality. Males were found to be more susceptible to DDT than females.

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